

GE Healthcare

Ettan DALTsix Electrophoresis System

User Manual



Ettan

Important user information

All users must read this entire manual to fully understand the safe use of Ettan DALTsix Electrophoresis System.

Ettan DALTsix Electrophoresis System is intended for laboratory use only, not for clinical or *in vitro* use, or for diagnostic purposes.

WARNING!



The WARNING! sign highlights instructions that must be followed to avoid personal injury. Do not proceed until all stated conditions are clearly understood and met.

WARNING! High Voltage



The High Voltage sign highlights an instruction that must be strictly followed in order to avoid contact with lethal high voltage. Be sure not to proceed until the instructions are clearly understood and all stated conditions are met.

CAUTION!

The CAUTION! sign highlights instructions that must be followed to avoid damage to the product or other equipment. Do not proceed until all stated conditions are met and clearly understood.

Note

The Note sign is used to indicate information important for trouble-free and optimal use of the product.

CE Certifying

This product meets all requirements of applicable CE-directives. A copy of the corresponding Declaration of Conformity is available on request.

The **CE** symbol and corresponding declaration of conformity, is valid for the instrument when it is:

- used as a stand-alone unit, or
- connected to other CE-marked GE Healthcare instruments, or
- connected to other products recommended or described in this manual, and
- used in the same state as it was delivered from GE Healthcare except for alterations described in this manual.

Recycling



This symbol indicates that the waste of electrical and electronic equipment must not be disposed as unsorted municipal waste and must be collected separately. Please contact an authorized representative of the manufacturer for information concerning the decommissioning of equipment.

Safety standards

This product meets the requirement of the Low Voltage Directive (LVD) 73/23/EEC through the following harmonized standards:

- EN61010-1
- IEC 61010-1
- CAN/CSA-C22.2 No. 61010-1
- UL61010-1

EMC

This device meets the requirements of the EMC Directive 89 336/EEC through the following harmonized standard:

- EN 55014

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1 System function and description

1.1 *Electrophoresis System function and description*

In 2-D electrophoresis, proteins are separated according to isoelectric point by isoelectric focusing, most reliably on Immobiline™ DryStrip immobilized pH gradient (IPG) gel strips using the IPGphor™ or Multiphor™ II IEF Systems. The second-dimension electrophoresis separates the proteins on the basis of their molecular mass using sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE). The Ettan™ DALTsix Electrophoresis System is designed to handle large second-dimension gels in a simple, efficient, and reproducible manner.

1.1.1 **Electrophoresis Unit**

The Ettan DALTsix Electrophoresis Unit accommodates up to six 25.5 × 20.5 cm slab gels, either 1 mm or 1.5 mm thick, in a common tank under identical conditions. A sample, focused in an IPG strip, is placed on the cathodic (upper) surface of a slab gel and sealed in place with agarose. Up to six gel cassettes are inserted into the electrophoresis unit and any unused slots are filled with blank cassette inserts. The Upper Buffer Chamber/Buffer Seal (UBC) is pushed down over the cassettes, holding the gel cassettes in slots flanked by a double rubber gasket.

Use EPS 601 Power Supply for power supply. If any other power supply is used it must have similar specifications as the EPS 601 Power Supply, such as a floating output, leakage current detection and be capable of handling up to 600 V, 400 mA, or 100 W.

A pump mounted under the lower buffer chamber circulates the buffer, pumping it up into the chamber on the right-hand side, between the cassettes, down the left side, and over the internal heat exchanger before returning to the pump. The pump starts when it is plugged in. For temperature control, the heat exchanger located in the bottom of the unit must be connected to a MultiTemp™ III or similar circulating water bath, that have earthed water pipe and low pressure water circulation.

1.1.2 **Gel Caster**

The Ettan DALTsix Gel Caster holds up to six 1-mm or 1.5-mm gel cassettes, with separator sheets, for casting homogenous or gradient gels. When desired, fewer gels can be cast at one time by using blank cassette inserts to occupy any unneeded volume. The removable faceplate and separator sheets simplify loading and unloading the casting unit. The groove in the back of the caster provides the channel through which the gel solution is poured into the caster unless gradients are being the cast by displacement through the lower tubing fitting.

Ettan DALTsix System components

- Six slot vertical slab electrophoresis unit
- Gel caster
- Gel casting cassettes
- Gradient maker

1 System function and description
1.1 Electrophoresis System function and description

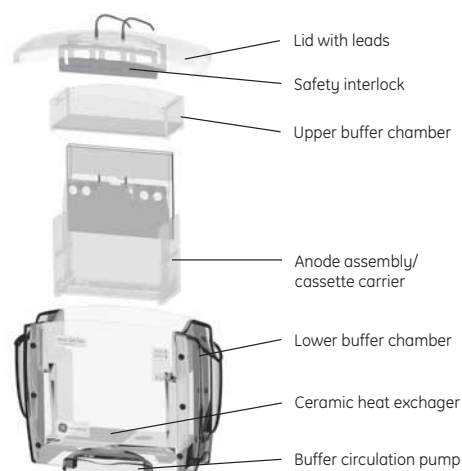


Fig 1-1. Exploded view of Ettan DALTsix Electrophoresis Unit.



Fig 1-2. Ettan DALTsix Gel Caster for casting 1.0 and 1.5 mm large format gels.

1.1.3 Gel Casting Cassettes

The Ettan DALTsix Electrophoresis System uses the same cassettes as the Ettan DALT*twelve* Electrophoresis System (see Fig 1-3). The gel casting cassettes are pre assembled. Two glass plates are joined along one edge by a hinge strip of silicone rubber, and the vinyl side spacers (1.0 mm or 1.5 mm thick) are glued in place. To complete assembly, close the two plates like a book and press the plates together over the length of the spacer. Gels are removed by opening the book after the run and carefully lifting out the gel slab. Care must be taken to ensure that the gel does not adhere to the spacers and tear during removal. The cassette is cleaned as a unit and can be stood upright to dry. The cassettes can be cleaned in an automatic dishwasher. Cassettes are 27 × 22 cm and produce a gel about 25.5 × 20.5 cm. A 1.0-mm thick gel has a volume of approximately 52 ml and a 1.5-mm thick gel has a volume of approximately 78 ml.



Fig 1-3. Gel Casting Cassettes.

1.1.4 Gradient Maker

The Ettan DALTsix Gradient Maker is designed for producing linear gradients of aqueous solutions ranging in volume from 200–2 000 ml. The gradient maker can be used to form convex and concave exponential gradients with the addition of a one-holed rubber stopper, a piece of rigid tubing, and a piece of flexible tubing.



Fig 1-4. Ettan DALTsix Gradient Maker.

1.2 Heat exchanger

The heat exchanger is built into the base of the separation tank. The white alumina ceramic heat exchange surface is fragile and the user should avoid dropping anything directly on surface.

When connected to a circulator bath, coolant passes through a serpentine chamber beneath the ceramic plate. The thin ceramic permits rapid heat exchange between the coolant and the buffer in the electrophoresis tank. The ceramic plate is attached with silicone rubber adhesive. The heat exchanger tubing connectors are 13-mm o.d.

The heat exchanger is rated to a maximum of 0.8 atmospheres above ambient (12 psig). Do not connect it to a water tap. Connect only to coolant sources with regulated pressure.

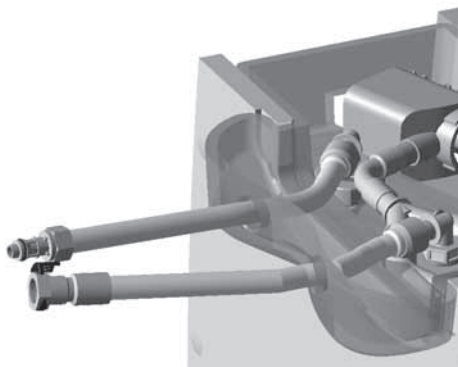


Fig 1-5. To connect the coolant tubing lines to the tank, remove the lid and the anode rack from the tank and lay the tank fill-label (front) side down. Guide the free ends of the coolant tubes through the holes in the tank base and slip them onto the plastic connector fittings, then pull the collector collar onto the tubing to lock it in place. The end of the tubing can be lubricated with glycerol or a mix of glycerol:water to ease the fit.

- 1 System function and description
- 1.2 Heat exchanger

2 Safety information

- This system is designed for indoor use only.
- To avoid risk of injury, operation and user maintenance should be performed by properly trained personnel only, and in accordance with the instructions.
- Any equipment connected to the system should meet the requirements of the EN 61 010-1 or other international safety standard. Within EU, connected equipment must be CE-labelled

2.1 Warnings



WARNING! Connect the instrument to a properly grounded electrical outlet.



WARNING! Use EPS 601 as power supply for Ettan DALTSix Electrophoresis System. If another power supply is used it must have similar specifications as EPS 601, such as floating output and leakage current detection.



WARNING! The safety lid must be firmly in place before power can be applied.



WARNING! *Always* turn off the power to the gels before opening the safety lid.



WARNING! Only connect MultiTemp III, or a circulating water bath) with earthed water pipe and low pressure water circulation. Do not connect the heat exchanger to a water tap or any other water source with unregulated pressure that may exceed 82 kPa (12 PSI).



WARNING! Check electrical cables for damage and the electrophoresis unit for damage or leakage before use. If damage and/or leakage is present, do NOT use the equipment. There is a risk for high voltage injuries.



WARNING! If electrical cables are damaged and/or any of the buffer chambers has a leakage do NOT use the equipment. There is a risk for high voltage injuries!



WARNING! The power cord should be easily accessible to enable emergency disconnection of the current.



WARNING! Never connect the Ettan DALTSix Electrophoresis System to higher voltage, current or effect than specified for that system.



WARNING! Always disconnect the power cord before draining the tank or before servicing the Ettan DALTSix Electrophoresis System.



WARNING! Use care when lifting and moving the electrophoresis unit. It is best to move the unit when empty.



WARNING! The casting unit, when filled with glass plates and gel solutions, is very heavy. Use caution when trying to move or lift the caster.



WARNING! Fire hazard, never use flammable liquids in the electrophoresis tank.



WARNING! The protection provided by the equipment may be impaired if this equipment is used in a manner not specified by the manufacturer.



WARNING! Only accessories and parts approved or supplied by GE Healthcare may be used for operating, maintaining and servicing this product.

2.2 Cautions

CAUTION! Rinse and flush the tank and pumping system with distilled or deionized water before and after use.

CAUTION! Do not run the circulation pump if the electrophoresis tank is empty.

CAUTION! Do not operate with buffer temperature above 40°C. All plastic parts are rated for 40°C continuous duty.

CAUTION! Turn the buffer circulation pump on during electrophoresis to minimize uneven heating even if not connected to a thermostatted circulator.

CAUTION! Connect the heat exchanger to an external thermostatted circulating bath. Overheating will cause irreparable damage to the unit.

CAUTION! Do not autoclave or boil this unit or any of its parts.

2.3 Emergency procedures

Emergency shutdown

In a situation where there is a risk of injury, turn off the current by:

- disconnecting the external power supply cable from its power outlet.
- disconnecting the Ettan DALTSix from its power outlet.


Power failure routine

In the event of a power failure, the run is interrupted in an undefined state.

2 Safety information
2.3 Emergency procedures

3 Specifications

3.1 Electrophoresis Unit

Maximum voltage (high voltage connection)	600 V 
Maximum current (high voltage connection)	400 mA
Maximum power (high voltage connection)	100 W
Gel capacity	6 gels
Electrophoresis buffer volume	5.5 l
Dimensions (h × w × d)	40.3 × 54.2 × 16.0 cm
Weight (empty)	11.1 kg
Weight (filled)	16.8 kg
Maximum temperature	40°C
Environmental operating conditions	Indoor use, 4–40°C Humidity up to 90% Altitude to 2000 m
Installation category	II
Pollution degree	2
100 V~ model	100 V~, 50-60 Hz, 8 W (80649477)
115 V~ model	110–120 V~, 60 Hz, 8 W (80648508)
230 V~ model	220–240 V~, 50-60 Hz, 7 W (80648527)
Compliance with standards	The declaration of conformity is valid for the instrument only if it is: <ul style="list-style-type: none"> • used in laboratory locations • used in the same state as it was delivered from GE Healthcare except for alterations described in the User Manual • connected to other CE labelled GE Healthcare modules or other products as recommended. Compliance with safety standards: <ul style="list-style-type: none"> • IEC61010-1 • EN61010-1 • UL61010-1 • CAN/CSA-C22.2 No. 61010-1

3.2 Gel Caster

Gel capacity	6 gels, 1.0 mm or 1.5 mm thick
Acrylamide solution volume (total)	425 ml for 1.0 mm thick gels, 550 ml for 1.5 mm thick gels
Dimensions (h × w × d)	27 × 37 × 8 cm
Weight (empty)	3.6 kg
1.0 mm Gel Casting Cassette	
Cassette dimensions (w × h × d)	27.6 × 21.7 × 0.70 cm
Slab gel dimensions (w × h × d)	25.5 × 20.5 × 0.10 cm
1.5 mm Gel Casting Cassette	
Cassette dimensions (w × h × d)	27.6 × 21.7 × 0.75 cm
Slab gel dimensions (w × h × d)	25.5 × 20.5 × 0.15 cm
Ettan DALTsix Gradient Maker	
Volume	1000 ml
Weight	0.73 kg

4 Preparing the Gel Caster

Set up the gel caster near a sink, in a tray, or on a drainboard so that any liquid that overflows, spills, or drains out of the unit during pouring or disassembly can be easily contained.

The Ettan DALTsix Gel Caster accommodates up to six 1-mm or 1.5-mm gel cassettes with separator sheets (0.5 mm) between them. If you are not planning to cast a full set of gels, use the blank cassette inserts (purchased separately) with separator sheets between them to occupy the extra space.

Gel labels, for easy indexing of gels and samples, can be placed in the cassettes at any time during the assembly of the caster.

- 1 Check that the caster is level. Remove the faceplate and lay the caster on its back. If casting gels by the displacement method, remove the triangular wedge in the V-shaped base.
- 2 Fill the gel caster starting with a separator sheet against the back wall. The separator sheets make it easier to remove the cassettes from the unit after polymerization. Fill the caster by alternating cassettes with separator sheets. The rubber hinge should be on the left side of the caster, with the matching ends of the cassette down. End with a separator sheet then use the thicker filler sheets (1.0 mm) to bring the level of the stack of cassettes and spacers even with the edge of the caster.
- 3 Remove the gray foam seal from the groove in the faceplate and lubricate it with a light coating of GelSeal compound to help ensure a liquid-tight seal. Place the gasket back in the groove on the faceplate. Avoid stretching the gasket by seating it from the ends first, working toward the middle.
- 4 Turn two black knobbed screws into the two threaded holes across the bottom until they are well engaged (two to three full turns). Carefully place the faceplate onto the caster with the bottom slots resting on their respective screws. Clamp both sides of the faceplate with six spring clips and tighten the screws. Be sure the sealing gasket is compressed evenly by the faceplate and forms a tight seal with the caster. Do not overtighten the screws.
- 5 If casting using the filling channel in the back plate of the caster, be sure to plug the barbed fitting in the faceplate. If casting using the displacement method, attach the tubing from the peristaltic pump to the barbed fitting.

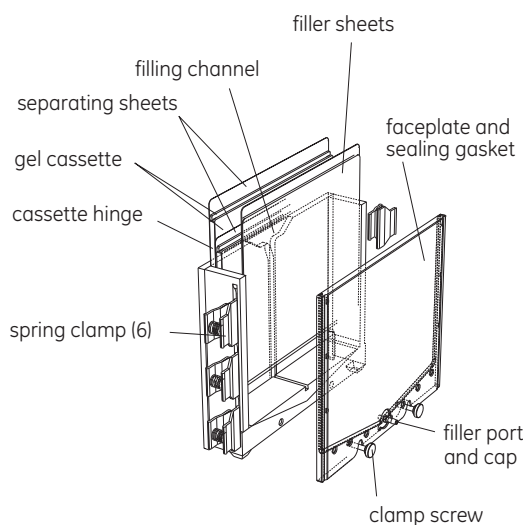


Fig 4-1. Gel caster with parts labelled.

5 Casting homogeneous gels



WARNING! When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the chemicals used. Follow local regulations and instructions for safe operation and maintenance of the system.



WARNING! Acrylamide is a neurotoxin. Never pipette by mouth and always wear protective gloves when working with acrylamide solutions, IPG strips, or surfaces that come into contact with acrylamide solutions.

- 1 Be sure the entire gel casting system is clean, dry, and free from any polymerized acrylamide.
- 2 Prepare a sufficient volume of gel overlay solution (water-saturated n-butanol). You need 1.0 ml of overlay for each 1-mm cassette and 1.5 ml for overlay for each 1.5-mm cassette. Mixing 1-mm and 1.5-mm cassettes, in one casting is not advised.

Note: If using glass plates in stead of cassettes, ensure that the plates are properly aligned.

- 3 If casting using the displacement method, make up 200 ml of displacing solution (0.375 M Tris-Cl pH 8.8, 30% (v/v) glycerol, bromophenol blue).
- 4 For a full 6-gel, 1.5-mm cassette set, make up 600 ml of acrylamide gel stock solution without ammonium persulfate (APS) or N,N,N',N'-tetramethylethylenediamine (TEMED). For a full 6-gel, 1-mm cassette set, make up 450 ml of acrylamide gel stock solution as above. This amount of gel solution will provide you with sufficient volume to cast gels using either the filling channel or a peristaltic pump.

Note: For best result, the acrylamide gel stock solution should be 8°C when APS and TEMED is added.

- 5 Assemble the gel caster as described in the proceeding section; the caster should be placed on a level bench or on a leveling table so that gel tops are level.
- 6 Add the appropriate volumes of APS and TEMED only when ready to pour the gels, not before. Once these two components are added, polymerization begins and the gel solution should be completely poured within 10 min.
- 7 Pour the gel solution into the filling channel. If displacement casting is being used, the flow rate on the peristaltic pump should be increased slowly to the desired speed to avoid introducing any air bubbles.
- 8 For displacement casting, pump the gel solution into the caster until it is about 7–11 cm below the final desired gel height. Stop the flow of acrylamide and transfer the feed tube to the container holding the displacement solution. Restart the pump to displace the gel solution in the tubing and the V-trough at the bottom of the caster. The remaining acrylamide solution is forced into the cassettes to the final gel height. When the final desired gel height is reached, stop the pump and clamp off the tubing near the barbed fitting at the bottom of the caster.

For filling channel casting, pour the gel solution into the channel until the solution reaches the final desired height, about 1 cm below the top edge of the short plate.

The amount of gel solution required in either case will be ~550 ml for six 1.5-mm gels and ~425 ml for six 1-mm gels.

- 9 Immediately pipette the water-saturated *n*-butanol onto each gel. Avoid wetting adjacent plastic surfaces of the caster with *n*-butanol.

OR

Spray (do not pipette) 0.1% (w/v) SDS solution on the edges of the cassettes, for example with a plant sprayer, so the gels are covered. By doing this curved edges on the gels are prevented.

- 10 Allow the homogeneous gels to polymerize for at least one hour before disassembling the caster. If the peristaltic pump outlet tubing was connected directly to the caster, it should not be removed until polymerization is complete. Do not leave the water-saturated *n*-butanol on the gels overnight as long term exposure of acrylic to butanol will damage the caster.

6 Casting gradient gels

6.1 Preparation for gradient gel casting

Successful gradient gel casting requires planning, timing, and practice. A full cast with the Ettan DALTsix Gel Caster requires 420–550 ml of acrylamide stock. Polymerization begins as soon as TEMED and APS are added to the acrylamide stock. At this point there is no time to adjust the gradient maker, or the cassettes and separators in the gel caster. To familiarize yourself with the gel caster and gradient maker before casting gels, you should set up the unit and carry out a practice gradient cast, substituting water for the appropriate volume of light solution, and a mixture of glycerol and water for the appropriate volume of heavy solution.

A pore gradient gel results from mixing varying proportions of two solutions of different acrylamide concentrations and densities, a light solution and a heavy solution. The heavy gel solution contains glycerol and a higher concentration of acrylamide. During the gradient pouring procedure, the mixing ratio of high concentration acrylamide solution to low concentration solution gradually changes, with the heavier (high concentration) solution sinking below the light solution. This generates a downward gradient of increasing acrylamide (gel) percentage.

6.2 Gradient casting setup



WARNING! When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the chemicals used. Follow local regulations and instructions for safe operation and maintenance of the system.



WARNING! Acrylamide is a neurotoxin. Never pipette by mouth and always wear protective gloves when working with acrylamide solutions, IPG strips, or surfaces that come into contact with acrylamide solutions.

- 1 Be sure the entire gel casting system is clean, dry, and free from any polymerized acrylamide. Remove the rubber insert from the base of the V-shaped feed channel of the caster. The caster should be placed on a level bench to ensure that the gels and gradients are even and level.
- 2 Add a magnetic stir bar of the appropriate dimensions (20–30 mm long) to the mixing chamber and place the unit on a magnetic stirrer. If volumes will be less than half the capacity of the unit, an identical stir bar should be placed in the reservoir chamber as well to balance the displacement and prevent backflow into the reservoir when the chambers are first connected.
- 3 Connect tubing to the outlet connector and pump, and adjust pump speed, if used. Connect the tubing to the gel casting unit.
- 4 Close the connecting valve between the reservoir (heavy) and mixing (light) chambers and clamp off the exit line.
- 5 Prepare a sufficient volume of gel overlay solution (water-saturated n-butanol). You need 1.0 ml of overlay for each 1-mm cassette and 1.5 ml for each 1.5-mm cassette.
- 6 Make up 200 ml of displacing solution.
- 7 Make up the gel acrylamide solutions from the stock mixes, but do not add the 10% ammonium persulfate (APS) and 10% N,N,N',N'-tetramethylethylenediamine (TEMED). See Section 6.3.

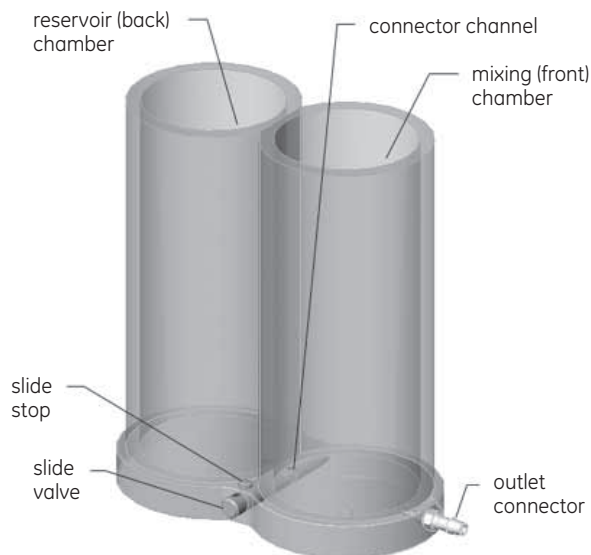


Fig 6-1. DALTsix gradient maker.

6.3 Pouring gel solutions for gradient gels

- 1 Prepare the gel caster, as described in Section 4, and place identifying gel labels inside each cassette.
- 2 When you are ready to cast the gels, add the APS and TEMED and mix each gel solution thoroughly. Vary the amount of TEMED added to control the rate of polymerization. Once these reagents are added, polymerization begins. You have about 10 min to cast the gradient before the gels begin to solidify at the top. Work rapidly and carefully.
- 3 Close the slide valve (out on side of white slide stop button, Fig 6-1). If the outlet tubing is not controlled by a pump, clamp it off near the gradient former. Add the required volume of the final solution to the reservoir (back) chamber.
- 4 Carefully open the slide valve (valve in on button side) and allow just enough solution to flow through the connector channel to fill it to the edge of the mixing chamber, then close the valve. Be sure no large bubbles remain to obstruct flow through the channel.
- 5 Add the required volume of the light solution to the mixing chamber and start the magnetic stirrer.
- 6 Simultaneously, open the outlet tubing if clamped off, and start the pump. Adjust the pump rate so that the solution is not forced up in a "fountain" that mixes with the overlying solution.
- 7 Immediately open the connecting valve between the mixing and the reservoir chamber.
- 8 Watch the gradient solution enter the caster. It is important that no bubbles disturb the gradient-watch the delivery carefully.
- 9 Just as the last of the gradient mix is pumped out of the mixing chamber, add 200 ml of displacement solution to the mixing chamber and pump it through until almost all of the gradient mix has entered the cassettes. It is convenient to include a dye in the displacement solution to visually track the boundary between the gradient mix and the displacement solution.
- 10 When the gels have reached the desired height and before air is introduced, stop the peristaltic pump. It is important that no air bubbles disturb the gradient.

- 11 Immediately pipette the water saturated *n*-butanol onto each gel. Avoid wetting adjacent plastic surfaces of the caster with *n*-butanol.
- 12 Allow the gradient gels to polymerize for 2 hours before disassembling the caster. Gradient gel polymerization should proceed from the top down. If the peristaltic pump outlet tubing was connected directly to the caster, it should not be removed until polymerization is complete. Do not leave the water-saturated *n*-butanol on the gels overnight as long term exposure of acrylic to butanol will damage the caster.

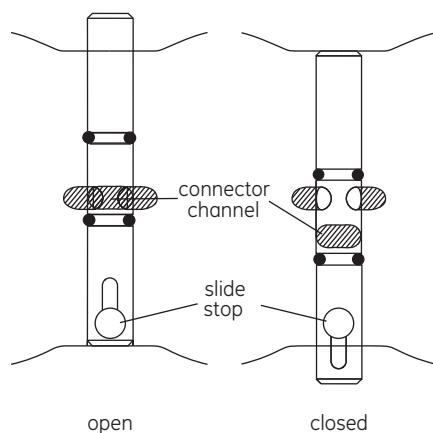


Fig 6-2. Open and closed positions of slide valve.

6 Casting gradient gels

6.3 Pouring gel solutions for gradient gels

7 Unloading the gel caster



WARNING! When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the chemicals used. Follow local regulations and instructions for safe operation and maintenance of the system.

- 1 Make sure the caster is either near a sink or on a tray so that any liquid leaking out can be contained.
- 2 Remove the front of the gel caster by loosening and removing the black-knobbed screws and spring clamps.
- 3 Carefully unload the cassettes from the unit by pulling forward on the separator sheets.
- 4 Rinse the top surface of each gel with distilled water to remove the n-butanol and any unpolymerized acrylamide. Remove the separator sheet if still attached and rinse the glass cassettes with water to remove any acrylamide adhering to the glass plates.
- 5 Examine the gels for polymerization defects and discard any unsatisfactory gels.
- 6 Store the acceptable gels in an airtight container at 4°C with a small amount of gel storage solution to keep the gels from drying out.
- 7 Rinse the gel caster and all tubing with mild detergent then rinse thoroughly with deionized water. Clean the separator and spacer sheets with a mild detergent and rinse with deionized water.

7 Unloading the gel caster

8 Electrophoresis

8.1 Placement of the electrophoresis unit

The Ettan DALTsix electrophoresis unit should be placed close to a sink for easy rinsing and draining. The tubing leading to and from the heat exchanger should be connected to a circulating water bath such as the MultiTemp III; the heat exchanger should not be connected to a water tap or any other coolant supply that lacks pressure regulation.

An EPS 601 or equivalent power supply should be placed conveniently near the electrophoresis unit in a dry place without risk for liquid exposure

8.2 Preparing second dimension gels.



WARNING! When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the chemicals used. Follow local regulations and instructions for safe operation and maintenance of the system.

8.2.1 Equilibration and loading

For a detailed description of the components of the SDS equilibration solution and the equilibration process, please consult *2-D Electrophoresis: Using Immobilized pH Gradients* (80-6429-60).

- 1 Prepare SDS equilibration buffer. Just prior to use, add DTT to the buffer to a concentration of 1% (w/v).
- 2 Place the IPG strips in individual tubes with the support film toward the wall.
- 3 Add 10–15 ml of the DTT-containing solution to each tube. Typically, two 18 cm strips can be equilibrated with 10 ml of buffer or two 24 cm strips can be equilibrated with 15 ml of buffer.
- 4 Incubate the strips for 10–15 min with gentle agitation. Do not over-equilibrate, as proteins can diffuse out of the strip during this step.
- 5 Second equilibration. Prepare SDS equilibration buffer with iodoacetamide added to 2.5% (w/v) and repeat steps 3 and 4.
- 6 Before equilibration is completed, prepare the gel cassettes for loading by rinsing the top of the gel with deionized water and draining. Before loading the IPG strips make sure that the gel surface and plates are dry.
- 7 Lay the prepared gel flat on a clean surface, short glass plate side up.
- 8 Using forceps, remove the equilibrated IPG strip from the equilibration solution and rinse with fresh SDS electrophoresis buffer.

8 Electrophoresis

8.2 Preparing second dimension gels.

- 9 Holding one end of the IPG strip with forceps, carefully draw it across the exposed top part of the long gel plate until the strip is completely on the glass plate and centered. Using a thin plastic spatula, ruler, or spacer push against the plastic backing of the IPG strip, not the gel itself, and slide the strip between the two glass plates and down into contact with the surface of the slab gel. The strip should just rest on the surface of the gel. Avoid trapping air bubbles between strip and the slab gel or piercing the second dimension gel with the strip. By convention, the acidic, or pointed, end of the IPG strip is on the left. The gel face of the strip should not touch the opposite glass plate. (See Fig 8-1).

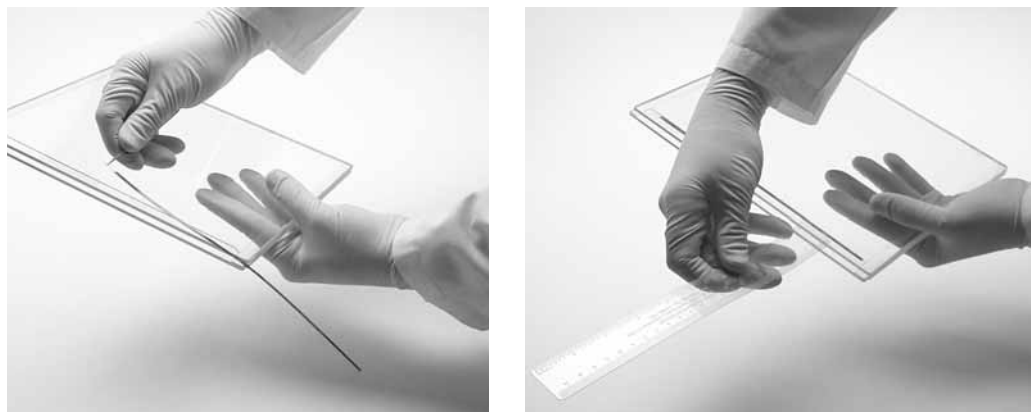


Fig 8-1. IPG strip are loaded onto cassette and slid into place.

- 10 Apply molecular weight marker proteins (optional). Apply the markers to a sample application piece in a volume of 15-20 μl then cover the piece with 50 μl of agarose sealing solution. Pick up the application piece with forceps and place next to one end of the IPG strip. The markers should contain 0.2-1.0 μg of each component for Coomassie blue staining and about 10-50 ng of each component for silver staining.
- 11 Seal the IPG strip in place. For each IPG strip, melt an aliquot of agarose sealing solution in a heating block or boiling water bath. (Tip: an ideal time to carry out this step is during IPG strip equilibration). Allow the agarose to cool slightly and slowly pipette the solution across the length of the IPG strip taking care not to introduce or trap bubbles. It will flow down between the glass plate and the support film and seal the IPG strip in place (see Fig 8-2). Agarose should also be used to seal any gap between the side of the gel and a side spacer. Allow a minimum of 1 min for the agarose to cool and solidify.



Fig 8-2. Adding agarose overlay.

8.3 Preparing the electrophoresis unit

Note: This Section describes the procedure for preparing the electrophoresis unit using the upper buffer chamber (UBC) shown in Fig 8-3 . If another UBC is used follow instructions supplied together with that UBC.



Fig 8-3. Upper chamber to be used for this instruction.

The Ettan DALTsix electrophoresis unit requires a total volume of about 5.7 l of electrophoresis buffer to fill both the UBC and the lower buffer chamber (LBC). For lab cast Laemmli gels, the LBC requires approximately 4.5 l of 1× SDS electrophoresis buffer while the UBC requires 1.2 l of buffer with a higher concentration. Prepare the required buffers as described below.

Note: Do not overfill the upper and lower chambers by using more buffer than required.

Electrophoresis buffer for Ettan DALTsix with lab-cast Laemmli gels

Gel thickness	Anodic buffer (LBC)	vol (l)	Cathodic buffer (UBC)	vol (l)
1.0 mm gels	1×SDS electrophoresis buffer	4.5	2× SDS electrophoresis buffer	1.2
1.5 mm gels	1×SDS electrophoresis buffer	4.3	3× SDS electrophoresis buffer	1.2

For the DALT Pre-cast Gels and Buffer Kit, only half of the 100× anode (lower) buffer is used for each run; the slightly reduced buffer concentration should not affect the run conditions. To prepare the anode buffer, dilute half the contents of the 100× anode buffer into 4.5 l of water. For the cathodic buffer, a single bottle of 10× cathode (upper) buffer should be diluted to a final volume of 1.2 l with deionized water and the full amount added to the UBC. Please consult the user instructions for the DALT Pre-cast Gels for detailed technical information regarding the assembly of gels in their cassettes.



WARNING! Check electrical cables for damage and the electrophoresis unit for damage or leakage before use. If damage and/or leakage is present, do NOT use the equipment. There is a risk for high voltage injuries.



WARNING! The high voltage current MUST always be disconnected when the safety lid of the electrophoresis unit is taken off. The high voltage current MUST never be switched on unless the safety lid is on the electrophoresis unit.



WARNING! Handle the gels with care. The edges of the glass plates of the gels are sharp.

- 1 Fill the electrophoresis unit with 4.5 liters of 1× electrophoresis buffer or diluted anode buffer (*when using Ettan DALTsix Pre-cast gels*) and turn the pump on (Fig 8-4 A).
- 2 Insert the anode assembly into the tank so that the circulation ports are properly aligned. The anode assembly is keyed so that it may only be inserted in one orientation and the bottom edge of the assembly should fit into the slot in the bottom of the tank (Fig 8-4 B).
- 3 Turn on the MultiTemp III, adjust the temperature to the desired setting.
- 4 Insert the prepared gels into the unit. Fill any unused slots with blank cassette inserts.
- 5 Wet the UBC sealings with 1× electrophoresis buffer or 0.1% SDS (immerse the sealings in solution or spray the sealings of the UBC using a plant sprayer) and apply an even pressure on the UBC to carefully slide it over the gel cassettes.
- 6 Fill the upper (cathode) chamber with 2× electrophoresis buffer or diluted cathode buffer to a level between the fill lines (about 1.2 liters).
- 7 Fill the LBC with 1× electrophoresis buffer or water (*when using Ettan DALTsix Pre-cast gels*) to the same level as the upper chamber (Fig 8-4 D).
- 8 Place the safety lid on the unit and begin electrophoresis.

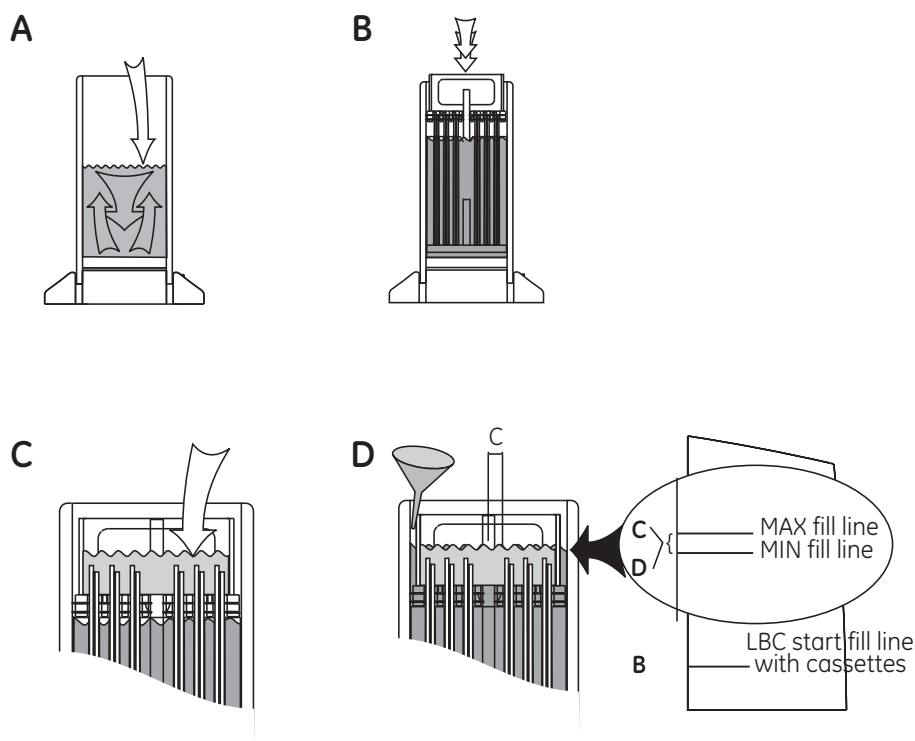


Fig 8-4. Filling the buffer chambers.

8.4 Recommended running conditions

Run conditions (Day run):

Applicable to 1 mm thick 12% PAA gel and Laemmli Buffer system. Set the MultiTemp temperature to 25°C.

Step	mA/gel	Voltage (V)	W/gel	Time (h:min)
1	10 ¹	80	1	1:00
2	40 ¹	500	13	4:30-6:00 ²

¹ For 1.5 mm gels, increase the current by 50%.

² Continue the electrophoresis until the bromophenol blue reaches the end of the gel.

Run Conditions (Overnight):

Set the MultiTemp temperature to 30°C.

Step	mA/gel	Voltage (V)	W/gel	Time (h)
1	10 ¹	80	1	1
2	12 ¹	150	2	15-17 ²

¹ For 1.5 mm gels, increase the current by 50%.

² Continue the electrophoresis until the bromophenol blue reaches the end of the gel.

8.5 Unloading the gels and cleaning the unit.



WARNING! The HIGH VOLTAGE and power switch MUST be turned off before removing the safety lid, unloading the gels, drain and cleaning the unit.

- 1 Carefully pull the UBC upward using the handles at each end.
- 2 As the UBC is pulled upward make sure that the cassettes remain in the anode assembly.
- 3 Take out the anode assembly.
- 4 Remove the gel cassettes from the anode assembly.
- 5 Open the gel cassettes using a Wonder Wedge (80-6127-88) to separate the plates on the side opposite the hinge.
- 6 For lab cast gels, run an edge of the Wonder Wedge down each side of the cassette along the spacer and carefully lift the gel out of the cassette. For pre-cast gels, lift the gel out by grasping the GelBond backing.
- 7 Tilt the entire electrophoresis unit on the side of the unit (do NOT tilt to front or back) to empty the buffer from the LBC into a sink or a waste container. See Section 8.1 for instructions of how to place the unit for safe use.



WARNING! Ettan DALTsix electrophoresis unit is heavy, especially when filled with buffer. Handle the unit with care to avoid personal damage.

- 8 Rinse all the components with distilled or deionized water. Flush the pump with distilled or deionized water by filling the unit and turning the pump on.

8 Electrophoresis

8.5 Unloading the gels and cleaning the unit.

9 Recipes



WARNING! When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the chemicals used.

Acrylamide stock (30.8%T)



WARNING! Acrylamide is a neurotoxin. Never pipette by mouth and always wear protective gloves when working with acrylamide solutions, IPG strips, or surfaces that come into contact with acrylamide solutions

	final conc.	amount
Acrylamide (MW 71.08)	30%	900 g
Bis (N,N'-methylenebisacrylamide, MW 154.17)	0.8%	24 g
Distilled or deionized water		to 3 000 ml

May need filtration. Weigh acrylamide and bis under a hood; avoid contact with dust. Filter and store at 4°C.

1.5 M Tris-Cl, pH 8.8

	final conc.	amount
Tris (MW 121.14)	1.5 M	545 g
6 M HCl to pH 8.8		about 150 ml
Distilled or deionized water		to 3 000 ml

Adjust to pH 8.8 and store at 4°C.

10% (w/v) SDS

	final conc.	amount
Sodium dodecylsulfate (MW 288.38)	10%	10 g
Distilled or deionized water		up to 100 ml

Store at room temperature.

10% (w/v) Ammonium persulfate

	final conc.	amount
Ammonium persulfate (MW 71.08)	10%	2 g
Distilled or deionized water		up to 20 ml

Prepare fresh.

10% (v/v) TEMED

	final conc.	amount
TEMED (MW 116.2)	10%	0.5 ml
Distilled or deionized water		4.5 ml

Prepare fresh.

Displacing solution

(0.375 M Tris-Cl, pH 8.8, 30% (v/v) glycerol, bromophenol blue, 200 ml)

	amount
Tris-Cl (1.5 M, pH 8.8)	50 ml
Glycerol	60 ml
Bromophenol blue	2 mg
Distilled or deionized water	90 ml

Should be made fresh; stored solution may support microbial growth.

Water-saturated butanol

	amount
n, i, or t-butanol	50 ml
Distilled or deionized water	10 ml

Combine in a bottle and shake. Use the top phase to overlay gels. Store at room temperature indefinitely.

Gel storage solution

(0.375 M Tris-Cl, pH 8.8, 0.1% (w/v) SDS, 2.0 l)

	final conc.	amount
Tris-Cl (1.5 M, pH 8.8)	0.375 M	500 ml
10% (w/v) SDS	0.1% (w/v)	20 ml
Distilled or deionized water		to 2 000 ml

Store at 4°C.

10× SDS electrophoresis buffer

(250 mM Tris, 1.92 M glycine, 1.0% (w/v) SDS, approximate pH 8.3, 20 l)

	final conc.	amount
Tris (MW 121.14)	250 mM	605.0 g
Glycine (MW 75.07)	1.92 M	2882.0 g
SDS (MW 288.38)	1.0% (w/v)	200.0 g
Distilled or deionized water		to 20 l

Do not adjust the pH of this solution.

SDS equilibration buffer

(50 mM Tris-Cl, pH 8.8, 6 M urea, 30% (v/v) glycerol, 2% (w/v) SDS, bromophenol blue, 200 ml)

	final conc.	amount
Tris-Cl (1.5 M, pH 8.8)	50 mM	6.7 ml
Urea (MW 60.06)	6 M	72.07 g
Glycerol (87% [v/v], MW 92.09)	30% (v/v)	69 ml
SDS (MW 288.38)	2% (w/v)	4.0 g
Bromophenol blue	trace	A few grains
Distilled or deionized water		to 200 ml

Store at -20°C. This is a stock solution. Add DTT or iodoacetamide before using.

Agarose sealing solution

(25 mM Tris, 192 mM glycine, 0.1% (w/v) SDS, bromophenol blue, 0.5% (w/v) agarose, 25 ml)

	final conc.	amount
1× SDS electrophoresis buffer (see above)		25 ml
Agarose (NA or M)		125 mg
Bromophenol blue	trace	A few grains

Combine all ingredients in a 250 ml Erlenmeyer flask. Swirl to disperse. On a low setting, heat in a microwave oven until the agarose is completely melted, about 1 min. Do not allow the solution to boil over. Allow the agarose to cool slightly before using. Do not adjust pH.

Homogeneous gel solutions**600 ml**

Final %	volume required for (ml)		
	10%	12.5%	15%
Acrylamide stock	200	250	300
1.5 M Tris-Cl, pH 8.8	150	150	150
Water	237	187	137
10% SDS	6.00	6.00	6.00
10% APS	6.00	6.00	6.00
10% TEMED	1.03	0.83	0.69

450 ml

final %T	volume required for (ml)		
	10%	12.5%	15%
Acrylamide stock	150	188	225
1.5 M Tris-Cl, pH 8.8	113	113	113
Water	178	140	103
10% SDS	4.5	4.5	4.5
10% APS	4.5	4.5	4.5
10% TEMED	0.77	0.62	0.52

Note: The amounts of TEMED (0.025-0.09% (v/v)) and APS (0.1% (w/v)) suggested here are based on our experience. You may want to change these volumes for your laboratory because of differences in temperature and reagent quality. Perform a small-scale test before using a new composition to check that your solution polymerizes in about 10 min. The gel recipes are based on Laemmli, U.K. Nature 227,680-685 (1970).

Gradient gel solutions

For six 1 mm gels:

Light solution, 210 ml

final %T	volume required for (ml)				
	8%	10%	12%	14%	16%
Acrylamide stock	55	68	82	95	109
1.5 M Tris-Cl, pH 8.8	52.5	52.5	52.5	52.5	52.5
Water	98	85	71	58	44
10% SDS	2.1	2.1	2.1	2.1	2.1
Glycerol	0	0	0	0	0
10% APS	2.1	2.1	2.1	2.1	2.1
10% TEMED	0.45	0.36	0.30	0.25	0.22

Heavy solution, 210 ml

final %T	volume required for (ml)				
	12%	14%	16%	18%	20%
Acrylamide stock	82	95	109	123	136
1.5 M Tris-Cl, pH 8.8	52.5	52.5	52.5	52.5	53
Water	55	41	27	14	0.0
10% SDS	2.1	2.1	2.1	2.1	2.1
Glycerol	18	18	18	18	18
10% APS	1.05	1.05	1.05	1.05	1.05
10% TEMED	0.09	0.08	0.07	0.06	0.06

For six 1.5 mm gels:**Light solution, 275 ml**

final %T	volume required for (ml)				
	8%	10%	12%	14%	16%
Acrylamide stock	73	92	110	128	147
1.5 M Tris-Cl, pH 8.8	69	69	69	69	69
Water	127	109	91	73	54
10% SDS	2.75	2.75	2.75	2.75	2.75
Glycerol	0	0	0	0	0
10% APS	2.75	2.75	2.75	2.75	2.75
10% TEMED	0.59	0.47	0.39	0.34	0.29

Heavy solution, 275 ml

final %T	volume required for (ml)				
	12%	14%	16%	18%	20%
Acrylamide stock	110	128	147	165	183
1.5 M Tris-Cl, pH 8.8	69	69	69	69	69
Water	73	55	37	18	0
10% SDS	2.75	2.75	2.75	2.75	2.75
Glycerol	19	19	19	19	19
10% APS	1.4	1.4	1.4	1.4	1.4
10% TEMED	0.13	0.11	0.10	0.09	0.08

10 Troubleshooting

10.1 Electrical and mechanical

Symptom	Possible causes	Possible solutions
No current at start of run	Insufficient volume of buffer in upper reservoir.	Ensure that the unit contains enough buffer to contact the upper electrode.
Buffer not circulating	Pump is not primed. Pump is off. Pump is broken.	Turn pump off and on to purge air bubbles. Turn on pump by plugging in the connector in the mains power outlet. Service call.

10.2 Gel casting

Symptom	Possible solutions
Gel caster leaks	Apply a light film of GelSeal compound to the foam gasket on the front plate before clamping and casting. Check the foam gasket for cracks or nicks and replace if necessary. If the stack is too thick, the front plate may not seat firmly against the gasket. Remove one or more of the filler sheets until the gasket seals.
Incomplete gel polymerization	Use only recent stocks of the highest quality reagents. If the dry ammonium persulfate does not crackle when water is added to it, replace with fresh reagent. Use fresh ammonium persulfate. Solutions of extreme pH may not polymerize. Degas the monomer solution. Oxygen inhibits polymerization. Increase both ammonium persulfate and TEMED by 30–50%. Adjust the gel solution temperature to a minimum of 20°C.
Gel is too soft, too brittle, or white	Check and adjust crosslinker concentration. Standard SDS gels should have a crosslinker concentration of 2.6% (%C = (g bis × 100)/(g monomer + g bis)). Make up fresh acrylamide stock solution.
Gel exhibits swirls	If gel polymerized too fast (<10 min), reduce the concentration of catalyst (APS and TEMED) by 25%. If gel polymerized too slowly (>50 min), increase the concentration of catalyst (APS and TEMED) by 50%. Make up fresh acrylamide stock solution.
Dye front curves up (smiles)	Check circulation of the buffer. Pre-chill the buffer. Decrease power, voltage, or current.
Vertical protein streaks	IPG strip not properly placed on gel surface. Make sure IPG strip contacts the gel surface uniformly along its entire length. Avoid gouging the surface of the separating gel. Include iodoacetamide equilibration step for IPG strip.
Gels cast simultaneously are different sizes	Allow the solution to settle, or reach equilibrium, before applying the overlay. Apply equal amounts of overlay solution to each gel. Apply overlay as quickly as possible.
Gradient gels-uneven layering	Add sucrose [15% (w/v)] or glycerol [25% (v/v)] to the high-percent monomer solution. Add a very small amount of bromophenol blue to the high-percent monomer solution to track gradient formation. Excessive bromophenol blue will inhibit polymerization.
Spots skewed or distorted	Gels run too fast – uneven migration. Run at a lower power setting. Use a two-step program: start at a low power setting until the proteins enter the gel, then increase the power for the remainder of the run. Uneven gel surface. Overlay the running gel with water-saturated butanol before polymerization begins to avoid forming an uneven gel surface. Uneven gel polymerization or gradient formation.
Heavy background after silver staining	Use reagents of the highest purity, preferably electrophoresis grade. Use deionized, double distilled water.
Unusually slow or fast run	Check for leaks. All plates, spacers, and gaskets must be clean, dry, and free of grease. Check the UBC. It should be free of nicks or tears. Check the pH of the buffer. If the pH is wrong, make fresh buffer, do not back-titrate. Check recipes, gel concentrations, and buffer dilutions. (For example, do not use Tris-HCl in place of Tris base for the electrophoresis buffer.) Discard older acrylamide solutions and use only reagents of highest quality. Only use freshly deionized urea of highest quality. Adjust power, current, or voltage.

10.3 Pre-cast gels

Symptom	Possible causes	Possible solutions
Second dimension electrophoresis proceeds slowly with high current	One of the slots in the upper buffer chamber is open The UBC is damaged. Anodic buffer has mixed with cathodic buffer from overfilling of either the cathodic or the anodic reservoir.	All 6 slots in the UBC should be occupied by either a gel cassette or a blank cassette. Carefully fill both buffer chambers to the same level. Ensure that the level of the anode (lower) buffer does not come above level of the buffer in the UBC when the electrophoresis unit is fully loaded.
Dye front is irregular	The top surface of the gel has been damaged during application of the IPG strip.	Take care during application of the IPG strip that neither gel is damaged.
	Bubbles between the gel and the glass plate.	Use the roller to remove any bubbles or excess liquid between the gel and the glass plate. Ensure that no visible bubbles remain and that the gel adheres firmly to the glass and resists movement.
	Liquid between the gel and the glass plate.	Ensure that no visible bubbles remain and that the gel adheres firmly to the glass and resists movement.
	Interfering substances in the first dimension.	Contaminants in the sample can cause distortions or swollen regions in the IPG strip following IEF. Modify sample preparation to limit these contaminants. See <i>2-D Electrophoresis Using Immobilized pH Gradients-Principles and Methods</i> (80-6429-60).
Pronounced downward curving of the dye front on one side of the gel	There is an unfilled gap between the gel and one of the spacers.	When sealing the IPG strip into place, ensure that some of the agarose sealing solution flows down any gap that may exist between the gel and spacer.
Distortion in the 2-D pattern	Bubbles between the gel and the glass plate.	Use the roller to remove any bubbles or excess liquid between the gel and the glass plate.
	Liquid between the gel and the glass plate.	Ensure that no visible bubbles remain and that the gel adheres firmly to the glass and resists movement.
	Interfering substances in the first dimension.	Contaminants in the sample can cause distortions or swollen regions in the IPG strip following IEF. These distortions can result in turn in disturbances in the second dimension.
Vertical gap in the 2-D pattern	Bubble between IPG strip and top surface of second dimension gel.	Ensure that no bubbles are trapped between the IPG strip and the top surface of second dimension gel.
Vertical streaking	Incorrectly prepared equilibration solution.	Prepare equilibration solution according to instructions.
	Poor transfer of protein from IPG strip to second dimension gel.	Use low power for sample entry phase. Extend entry phase if necessary.
	Insufficient equilibration	Extend equilibration time.
Spots are vertically doubled, or "twinned"	IPG strip is not placed properly.	Ensure that the plastic backing of the IPG strip is against the glass plate of the second dimension cassette.
Poor representation of higher molecular weight proteins	Incorrectly prepared equilibration solution.	Prepare equilibration solution according to instructions.
	Poor transfer of protein from IPG strip to second dimension gel.	Use low power for sample entry phase. Extend entry phase if necessary.

10.4 Stained gels

Symptom	Possible solution
Protein spots are diffuse or broader than usual	<p>Use only highest quality reagents.</p> <p>Make sure that polymerization is complete.</p> <p>Check equilibration time of IPG strips. Too long can lead to diffusion and too short can lead to incomplete equilibration.</p> <p>Make sure the IPG strip rests on the slab gel surface without damaging it.</p> <p>Problems with first dimension-see troubleshooting guides for IPGphor or Multiphor units, or 2-D Electrophoresis: Principles and Methods.</p>
Protein spots are poorly resolved	<p>Allow gel to polymerize completely.</p> <p>Begin electrophoresis as soon as the IPG strips are loaded to prevent diffusion of low molecular weight proteins.</p> <p>Running too fast. Reduce the power, current, or voltage.</p> <p>Reduce the temperature setting.</p> <p>Problems with the first dimension.</p>
Smeared or comet shaped spots	<p>Check pH of cathode buffer. Should be between 8.3 and 8.8.</p> <p>Make sure that 2× Laemmli buffer is used in the upper (cathode) chamber.</p> <p>Buffer or SDS depleted.</p>
Protein spots are near the buffer front	<p>Buffer depleted. Check pH of upper (cathode) buffer. Should be below pH 8.3-8.8. Be sure that 2× Laemmli buffer is being used in the upper (cathode) chamber.</p> <p>Pore size of the gel is too large. Increase the %T.</p> <p>Proteins degraded during sample preparation. Add protease inhibitors during sample preparation.</p> <p>Check the pH of the 4× gel buffer. It should be pH 8.8. Proteins will migrate faster below pH 8.8.</p>
Protein spots have not entered the gel when buffer front has reached the bottom of the gel	<p>The gel pore size is too small. Decrease the %T.</p> <p>Check the pH of the 4× gel buffer. It should be pH 8.8. Proteins will migrate slower above pH 8.8.</p>
Protein spots are at both extremes but not in center	<p>The molecular weight range of the sample requires an acrylamide concentration gradient to resolve the full range of proteins.</p>

10 Troubleshooting
10.4 Stained gels

11 Care and maintenance

11.1 Unpacking, inventory, and set-up

Unwrap all packages carefully and compare contents with the packing list, making sure all items arrived. If any part is missing, contact your local sales office. Inspect all components for damage that may have occurred while the unit was in transit. If any part appears damaged, contact the carrier immediately. Be sure to keep all packing material for damage claims or to use should it become necessary to return the unit.

11.2 Cleaning



WARNING! When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the chemicals used. Follow local regulations and instructions for safe operation and maintenance of the system.

For day-to-day operation of the unit, the cleaning procedure outlined in unit operation is sufficient, thoroughly rinsing the electrophoresis tank with distilled or deionized water. If desired the unit can be periodically cleaned with a dilute solution of a mild detergent.

Clean the Gel Casting Cassettes and Pre-cast Gel Cassettes with a dilute solution of a laboratory cleanser such as RBS-35, from Pierce Chemical Company. Rinse the cassettes thoroughly with distilled or deionized water.

- Do not autoclave or heat any part above 40°C.
- Do not expose the unit or its parts to organic solvents, including >20% ethanol.
- If using radioactive reagents, decontaminate the unit with a cleaning agent such as CONTRAD 70 or Decon 90 from Decon Laboratories, Inc.

Note: Local security regulations must be followed when handling radioactive reagents.

11.3 Replacement of components



WARNING! Never use the equipment if any part cracked or broken. Risk for electric shock

If any of the components becomes cracked or broken it should be replaced. The upper buffer chamber (UBC) should be replaced if any of the rubber flaps become cut or torn. To prevent damage to the flaps, ensure that all sharp edges on glass plates are smoothed and exercise care when employing the cassette removal tool. With normal use the UBC ribs may become bowed upward; this bowing should not affect the performance of the seal.

11.4 Recycling

Ettan Spot Handling Workstation contains the following materials:

- Stainless steel
- Aluminium
- Glass
- Plastic:
 - Acetal (POM)
 - Polyethylene Terephthalate Polyester (PETP)

11 Care and maintenance

11.5 Maintenance

- Polyurethane (PUR)

- Electrical and electronic components

When taking Ettan Spot Handling Workstation out of service these different materials must be separated and recycled according to local regulations.



This symbol indicates that the waste of electrical and electronic equipment must not be disposed as unsorted municipal waste and must be collected separately. Please contact an authorized representative of the manufacturer for information concerning the decommissioning of your equipment.

11.5 Maintenance

Check the following before each run on Ettan DALTsix:

- all parts are clean, see Section 11.2.
- all parts are intact and free of cracks and other damage. If a part is damaged replace the part or entire instrument before use, see Section 11.3.
- the power cord and high voltage cable is intact and free of cracks and other damage. If the cable is damaged replace it before use.
- there is no leakage from parts containing buffer or cooling water. Moist or liquid must be removed from the equipment before use.
- the circulation pump spins and that there is circulation of liquid.

12 Customer service information

12.1 Technical service and repair

GE Healthcare offers complete technical support for all our products. If you have any questions about how to use this product, or would like to arrange to repair it, please contact your local GE Healthcare representative for more information.

Note: Only use spare parts that GE Healthcare recommends for safety reasons. The warranty is not valid if parts that are not recommended by GE Healthcare are used.

12.2 Ordering information

Product	code no.
Ettan DALTsix Electrophoresis Unit	
100 V~	80-6494-77
110-120 V~	80-6485-08
220-240 V~	80-6485-27
Replacement Lid	80-6490-40
Replacement Upper Buffer Chamber	80-6490-78
Replacement Anode Assembly/Cassette Carrier	80-6491-35
Ettan DALTsix and DALT II Cassettes	
Pre-Cast Gel Cassette	80-6466-65
Gel Casting Cassette, 1-mm	80-6466-84
Gel Casting Cassette, 1.5-mm	80-6488-69
Blank Cassette Insert	80-6467-03
Cassette Removal Tool (2/pkg)	80-6474-82
Ettan DALT II Pre-Cast Gels	
Pre-Cast Gel 12.5% (6/pkg)	17-6002-36
Buffer Kit (one run of 12 gels)	17-6002-50
Ettan DALTsix Gel Caster	
Complete with Separator Sheets (7 pcs) and Filler Sheets (6 pcs)	80-6485-46
Rubber Insert	80-6493-44
Small Black Knobbed Screws	80-6493-63
Ettan DALTsix Gradient Maker	80-6487-36
Ettan DALTtwelve Gel Caster	
Complete with Separator Sheets (16 pcs) and Filler Sheets (6 pcs)	80-6467-22
Separator Sheets (16/pkg)	80-6467-41
Filler Sheets (6/pkg)	80-6467-60
Black-Knobbed Screws (4/pkg)	80-6437-58
Funnel Sponge	80-6474-06
Acrylic Feed Tube	80-6437-20
Foam Sealing Gasket	80-6023-76
Silicon Tubing Set, two pieces/pkg: 9 mm o.d., 178 mm long and 12.5 mm o.d., 16 mm long	80-6437-39
Replacement Tilt Leg with Nylon Screw	80-6474-25
Replacement Face Plate	80-6474-44

12 Customer service information

12.2 Ordering information

Product	code no.
Accessories	
2-D Electrophoresis: Using Immobilized pH Gradients	80-6429-60
Cassette Rack (2/pkg)	80-6467-98
Equilibration Tubes (12/pkg)	80-6467-79
Stainless Steel Staining Tray Set	80-6468-17
GelSeal, 1/4 oz. tube	80-6421-43
Roller	80-1106-79
Fluorescent Rulers (2/pkg)	80-6223-83
Wonder Wedge	80-6127-88
PlusOne Electrophoresis Chemicals and Reagents	
Urea, 500 g	17-1319-01
Dithiothreitol (DTT), 1 g	17-1318-01
Bromophenol Blue, 10 g	17-1329-01
Glycerol (87%), 1 l	17-1325-01
Acrylamide IEF (acrylic acid <0.002%), 1 kg	17-1300-02
Acrylamide IEF 40% solution, 1 l	17-1301-01
N,N',-Methylene bisacrylamide, 25 g	17-1304-01
N,N',-Methylene bisacrylamide 2% solution, 1 l	17-1306-01
Agarose NA, 10 g	17-0422-01
N,N,N',N', -tetramethylethylenediamine (TEMED), 25 ml	17-1312-01
Ammonium Persulfate (APS), 25 g	17-1311-01
Tris, 500 g	17-1321-01
Glycine, 500 g	17-1323-01
Sodium Dodecylsulfate (SDS), 100 g	17-1313-01
Silver Staining Kit, Protein	17-1150-01
Molecular Weight Markers	
MW Range 2512-16949, 2 mg/vial, 1 vial	80-1129-83
MW Range 14400-94000, 575 (mg/vial, 10 vials	17-0446-01
MW Range 53000-212000, 175 (mg/vial, 10 vials	17-0615-01
Companion Products	
EPS 601 Power Supply 6 - 600 V, 1- 400 mA, 1-100 W	18-1130-02
MultiTemp III Thermostatic Circulator, 230 V	18-1102-78
MultiTemp III Thermostatic Circulator, 115 V	18-1102-77

www.gelifesciences.com

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